Twisted-wing parasites feel no pain

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Jena PhD student and first author of the study Kenny Jandausch with an experimental set-up for twisted-wing parasites. Credit: University of Jena

Reproduction in the insect order Strepsiptera—also known as twisted-wing parasites—is not for the fainthearted. To inseminate the eggs of its partner, the male partner injures the "neck" of the female with its hook-shaped penis and injects the seminal fluid directly into the body cavity.

This traumatic insemination is risky for the female. For example, the injury may result in a loss of body fluid and invading germs may cause infections.

However, in the course of evolution, the females of the strepsipteran species Stylops ovinae and Xenos vesparum have become morphologically well adapted to the brutal advances of their partners. This is the conclusion of a research team working at the universities of Jena, Kiel and Freiburg, and the Karlsruhe Institute of Technology, which reports on its findings currently in the scientific journal *PeerJ*.

Twisted-wing parasites can be found all over the world. The fact that almost no one knows them is probably because, like many other insects, they are rather small and inconspicuous, measuring only a few millimeters. Apart from the extremely short life span of the males—only a few hours—the decisive factor is probably that "the females of the vast majority of the species spend their entire lives as parasites well hidden in the abdomens of other insects," explains Dr. Hans Pohl from the University of Jena.

Only the anterior part of the female's body is available

Stylops ovinae, for example, lives in the mining bee (Andrena vaga) and Xenos vesparum in paper wasps (species of the genus Polistes). Only the cephalothorax, which is about as large as a pinhead, protrudes from the host. "So, in order to reproduce at all, the parasites have to come up with something," says Pohl, an insect expert and head of the team of authors of the present publication. Because the male cannot reach the usual mating area on the female abdomen, only the anterior part of the female's body is available to inject the sperm.

However, as the team has now discovered, the females of the two species studied are not defenseless against the males during the traumatic penetration. "We were able to show that the cuticle of the females of Stylops ovinae and Xenos vesparum is clearly thickened in a certain part of the body between the head and the trunk. This is the region where the male stabs the female with his penis," explains Ph.D. student Kenny Jandausch, first author of the study. The entire cuticle contains a lot of resilin, a protein molecule that makes the outer skin particularly elastic. However, as the skin at the puncture site is thicker than at other areas, the injury there is less dangerous for the female parasite because it allows the wound to close very efficiently. "In contrast to Xenos vesparum, this site forms a kind of pocket in Stylops ovinae into which the penis is inserted," says Jandausch.

Broad range of methods reveals mating mechanism

But how exactly do the females attract the males? First of all, in a very classical way: "Virgin females emit an odor that attracts males from the surrounding area," says Hans Pohl. The entomologists were able to observe this directly in the countryside around Jena. With a small cage of female twisted-wing parasites ready for mating, the researchers attracted males and took them back to their laboratory. There, they joined males and females in Petri dishes, observing them through a microscope.

"It turned out, firstly, that the females not only attracted males of Stylops ovinae, but also males of two other species of this genus. Secondly, it was observed that the males of the other two species were unable to insert their penis into the female's mating pocket despite vigorous efforts," says Kenny Jandausch.

Using computer tomography images, the researchers were able to show the morphological fit of penis and mating pocket and produced a three-

dimensional computer model. "In the process, we found out that only males of the same species are actually able to mate successfully with females," explains Pohl. "Our hypothesis is that the mating pocket represents a prezygotic barrier that prevents mating between different species before fertilization." The researchers had previously assumed that the odor emitted by female twisted-wing parasites only attract males of the same species.

In the end, however, there is bad news for female twisted-wing parasites: After the sperm has fertilized the thousands of eggs in her body, just as many tiny larvae develop and are born alive a few weeks later, an event that the mother herself does not survive.

More information: Kenny Jandausch et al, Have female twisted-wing parasites (Insecta: Strepsiptera) evolved tolerance traits as response to traumatic penetration?, *PeerJ* (2022). DOI: 10.7717/peerj.13655

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